We claim:

1. A non-natural cis-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, a loop I, and a loop II, wherein loop I and loop II are derived from loop I and loop II of a first hammerhead ribozyme selected from cherry small circular RNA+ (Scc+), cherry small circular RNA- (Scc-), Lucerne transient streak virusoid+ (sLTSV+), Lucerne transient streak virusoid- (sLTSV-), Tobacco ringspot virus satellite RNA+ (sTRSV+), Arabis mosaic virus (sArMV), Chicory yellow mottle virus satellite RNA (sCYMV), Barley yellow dwarf virus satellite RNA- (sBYDV-), Barley yellow dwarf virus satellite RNA+ (sBYDV+), Peach latent mosaic virus RNA+ (PLMVd+), Peach latent mosaic virus RNA- (PLMVd-), Chrysanthemum chlorotic mottle viroid+ (CChMVd+), Chrysanthemum chlorotic mottle viroid- (CChMVd-), Subterraneum clover mottle virusoid (vSCMoV), and velvet tobacco mottle virusoid (vVTMoV), and wherein at least one of stem I, stem II, and stem III is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

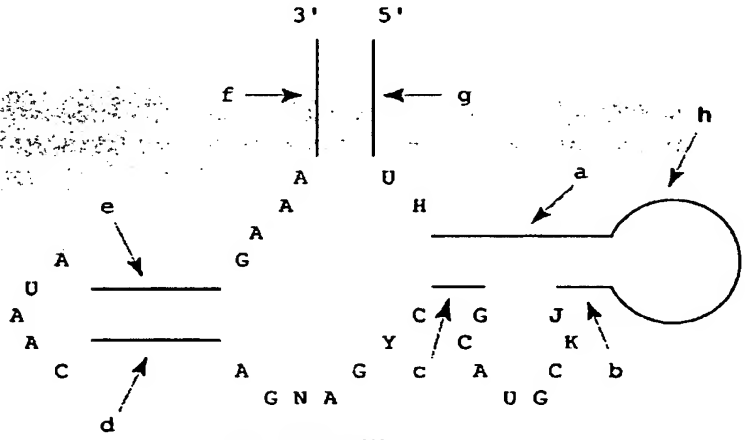
2. A non-natural cis-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, a bulge within stem I, and a loop II, wherein loop II and the bulge within stem I are derived from loop II and a bulge within stem I of a first hammerhead ribozyme selected from *Notophthalmus viridescens* satellite RNA (newt), *Ambystoma talpoideum* (Am. ta.), *Amphiuma tridactylum* (Am. tr.), *Schistosoma mansoni* hammerhead ribozyme (Schistozyme), *D. baccettii* cricket hammerhead ribozyme (cricketzyme A), *D. schiavazzii* cricket hammerhead ribozyme (cricketzyme B), and Avocado sunblotch viroid+ (ASBV+), and wherein at least one of stem II, stem III, and a portion of stem I is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

3. A non-natural cis-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, loop I, and a bulge within stem II, wherein loop I and the bulge within stem II are derived from a first ribozyme selected

from Chrysanthemum chlorotic mottle viroid- (CChMVd-) and Barley yellow dwarf virus satellite RNA (sBYDV+), and wherein at least one of stem I, stem III, and a portion of stem II is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

4. A non-natural cis-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, a bulge within stem I, and a bulge within stem II, wherein the bulge within stem I and the bulge within stem II are derived from a first ribozyme selected from Avocado sunblotch viroid- (ASBV-) and Carnation small viroid-like RNA+ (CarSV+), and wherein at least one of stem III, a portion of stem I, and a portion of stem II is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

5. A non-natural cis-cleaving hammerhead ribozyme comprising the structure III:



structure III

wherein:

J, K, and N are each independently selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

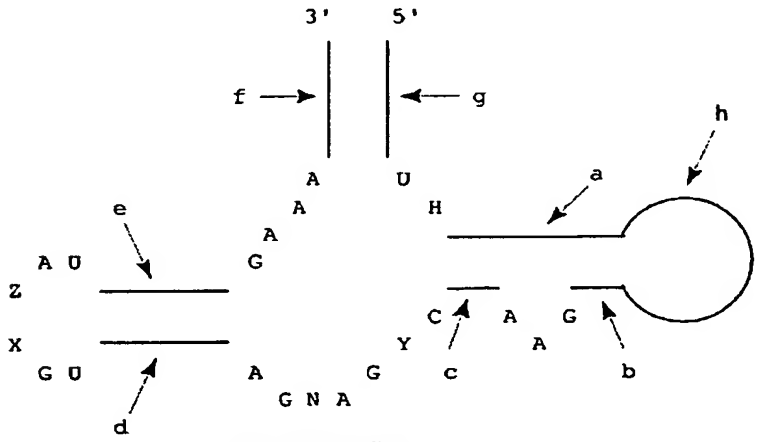
h is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, JKCGUACG, and c together are stem I;

d and e together are stem II; and

f and g together are at least a portion of stem III.

6. A non-natural cis-cleaving hammerhead ribozyme comprising the structure IV:



structure IV

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

X is selected from G and C;

Z is selected from G and A;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

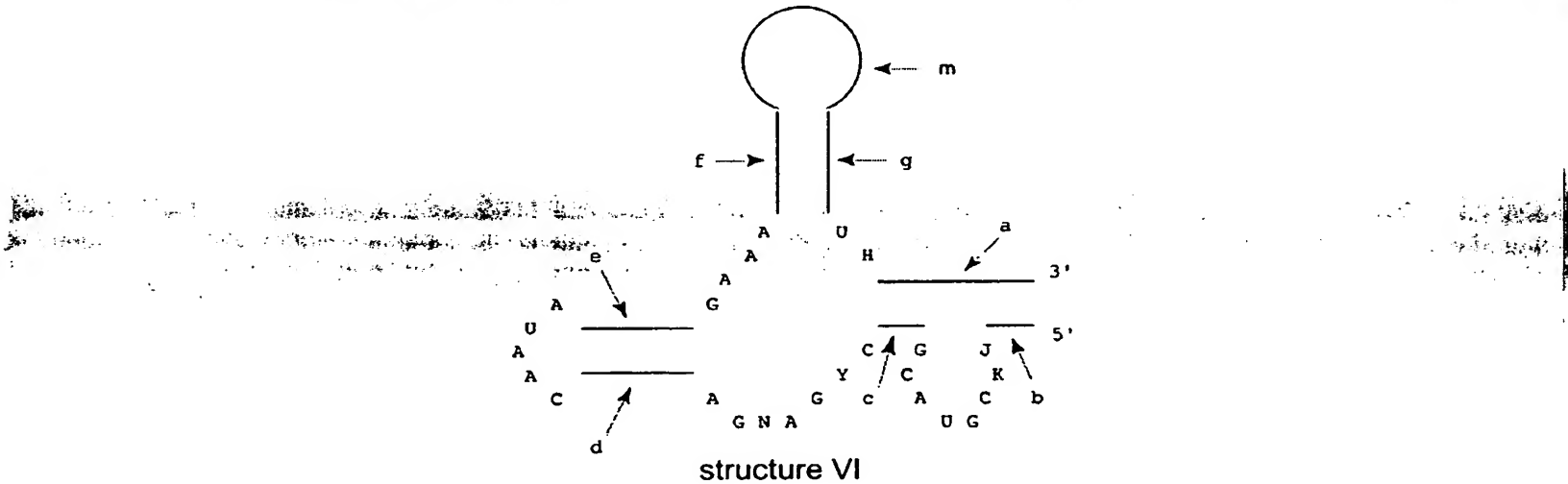
h is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, GAA, and c together are stem I;

d and e together are stem II; and

f and g together are stem III.

7. A non-natural cis-cleaving hammerhead ribozyme comprising the structure VI:



structure VI

wherein:

J, K, and N are each independently selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

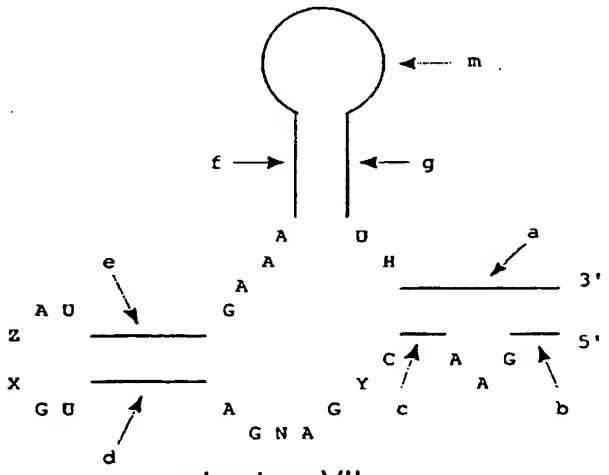
m is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, JKCGUACG, and c together are stem I;

d and e together are stem II; and

f and g together are at least a portion of stem III.

8. A non-natural cis-cleaving hammerhead ribozyme comprising the structure VII:



structure VII

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

X is selected from G and C;

Z is selected from G and A;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

m is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, GAA, and c together are stem I;

d and e together are stem II; and

f and g together are stem III.

9. The non-natural cis-cleaving hammerhead ribozyme of any of claims 1-8, wherein the ribozyme cis-cleaves at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 1 mM Mg^{2+} at 37°C.

10. The non-natural cis-cleaving hammerhead ribozyme of any of claims 1-8, wherein the ribozyme cis-cleaves at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 0.5 mM Mg^{2+} at 37°C.

11. The non-natural cis-cleaving hammerhead ribozyme of any of claims 1-8, wherein the ribozyme cis-cleaves at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 0.1 mM Mg^{2+} at 37°C.

12. A polynucleotide comprising a first nucleic acid sequence, wherein the first nucleic acid sequence encodes the non-natural cis-cleaving hammerhead ribozyme of any of claims 1-8.

13. The polynucleotide of claim 12, further comprising a second nucleic acid sequence, wherein the second nucleic acid sequence encodes an RNA that is not a non-natural hammerhead ribozyme.

14. The polynucleotide of claim 13, wherein the first nucleic acid sequence is inserted in frame into the second nucleic acid sequence.

15. The polynucleotide of claim 13, wherein the second nucleic acid sequence comprises a non-coding region, wherein the non-coding region is selected from a 3'-untranslated region (3'-UTR), a 5'-untranslated region (5'-

UTR), and an intron, and wherein the first nucleic acid sequence is inserted into the non-coding region.

16. A vector comprising the polynucleotide of claim 14.

17. A host cell comprising the polynucleotide of claim 16.

18. A vector comprising the polynucleotide of claim 15.

19. A host cell comprising with the polynucleotide of claim 18.

20. The non-natural hammerhead ribozyme of any of claims 5-8, wherein at least a portion of f and g together comprise an aptamer capable of binding a small molecule.

21. A polynucleotide comprising a first nucleic acid sequence, wherein the first nucleic acid sequence encodes the non-natural cis-cleaving hammerhead ribozyme of claim 20.

22. The polynucleotide of claim 21, further comprising a second nucleic acid sequence, wherein the second nucleic acid sequence encodes an RNA that is not a non-natural hammerhead ribozyme.

23. The polynucleotide of claim 22, wherein the first nucleic acid sequence is inserted in frame into the second nucleic acid sequence.

24. The polynucleotide of claim 22, wherein the second nucleic acid sequence comprises a non-coding region, wherein the non-coding region is selected from a 3'-untranslated region (3'-UTR), a 5'-untranslated region (5'-UTR), and an intron, and wherein the first nucleic acid sequence is inserted into the non-coding region.

25. A vector comprising the polynucleotide of claim 23.

26. A host cell comprising with the polynucleotide of claim 25.

27. A vector comprising the polynucleotide of claim 24.

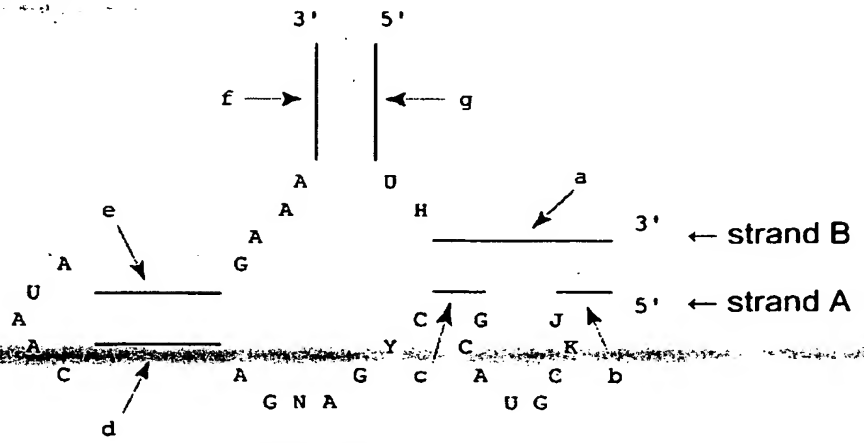
28. A host cell comprising with the polynucleotide of claim 27.

29. A non-natural trans-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, a bulge within stem I, and a loop II, wherein loop II and the bulge within stem I are derived from loop II and a bulge within stem I of a first hammerhead ribozyme selected from

Notophthalmus viridescens satellite RNA (newt), *Ambystoma talpoideum* (Am. ta.), *Amphiuma tridactylum* (Am. tr.), *Schistosoma mansoni* hammerhead ribozyme (Schistozyme), *D. baccettii* cricket hammerhead ribozyme (cricketzyme A), *D. schiavazzii* cricket hammerhead ribozyme (cricketzyme B), and Avocado sunblotch viroid+ (ASBV+), and wherein at least one of stem II, stem III, and a portion of stem I is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

30. A non-natural trans-cleaving hammerhead ribozyme comprising a core, a stem I, a stem II, a stem III, a bulge within stem I, and a bulge within stem II, wherein the bulge within stem I and the bulge within stem II are derived from a first ribozyme selected from Avocado sunblotch viroid- (ASBV-) and Carnation small viroid-like RNA+ (CarSV+), and wherein at least one of stem III, a portion of stem I, and a portion of stem II is derived from a second hammerhead ribozyme that is not the same as the first hammerhead ribozyme.

31. A non-natural trans-cleaving hammerhead ribozyme comprising strand A of structure I:



structure I

wherein the ribozyme cleaves a target RNA sequence comprising strand B of structure I;

wherein:

J, K, and N are each independently selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

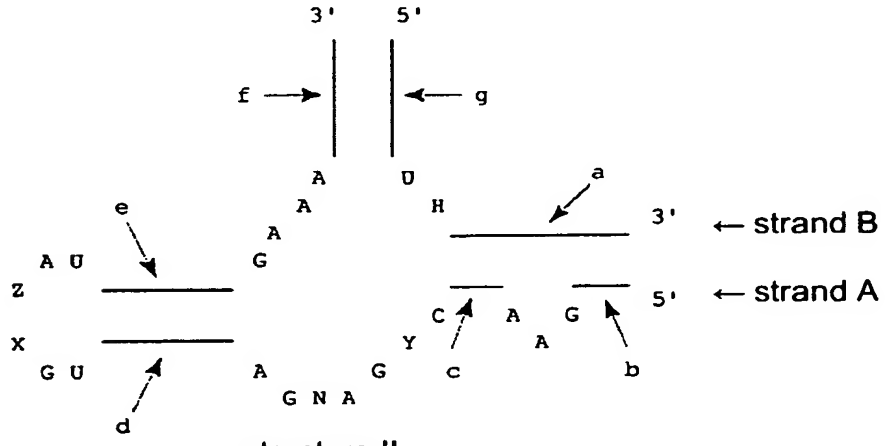
b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, JKCGUACG, and c together are stem I;

d and e together are stem II; and

f and g together are at least a portion of stem III.

32. A non-natural trans-cleaving hammerhead ribozyme comprising strand A of structure II:



structure II

wherein the ribozyme cleaves a target RNA sequence comprising strand B of structure II;

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

X is selected from G and C;

Z is selected from G and A;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, GAA, and c together are stem I;

d and e together are stem II; and

f and g together are stem III.

33. The non-natural trans-cleaving hammerhead ribozyme of any of claims 29-32, wherein the ribozyme cleaves the target RNA sequence at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 1 mM Mg^{2+} at 37°C.

34. The non-natural trans-cleaving hammerhead ribozyme of any of claims 29-32, wherein the ribozyme cleaves the target RNA sequence at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 0.5 mM Mg^{2+} at 37°C.

35. The non-natural trans-cleaving hammerhead ribozyme of any of claims 29-32, wherein the ribozyme cleaves the target RNA sequence at an initial rate of at least 0.5 min^{-1} in a buffer comprising 50 mM Tris (pH 7.0) and 0.1 mM Mg^{2+} at 37°C.

36. A polynucleotide comprising a nucleic acid encoding the non-natural trans-cleaving hammerhead ribozyme of any of claims 29-32.

37. A vector comprising the polynucleotide of claim 36.

38. A host cell comprising the polynucleotide of claim 36.

39. A pharmaceutical composition comprising the polynucleotide of claim 36 in a pharmaceutically acceptable carrier.

40. A pharmaceutical composition comprising the non-natural trans-cleaving hammerhead ribozyme of any of claims 29-32 in a pharmaceutically acceptable carrier.

41. A method of cleaving a target RNA in a mammal comprising administering the pharmaceutical composition of claim 39, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

42. A method of cleaving a target RNA in a mammal comprising administering the pharmaceutical composition of claim 40, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

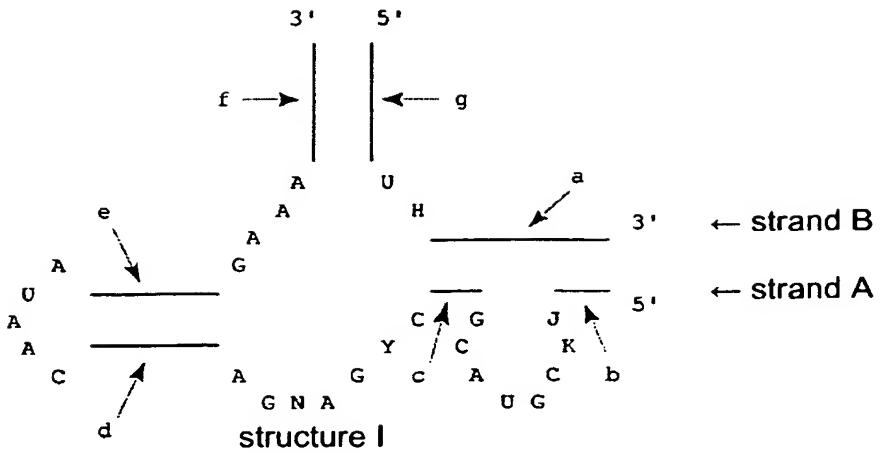
43. A method of decreasing the level of a target RNA in a mammal comprising administering the pharmaceutical composition of claim 39, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

44. A method of decreasing the level of a target RNA in a mammal comprising administering the pharmaceutical composition of claim 40, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

45. A method of decreasing the expression of a protein encoded by a target RNA in a mammal comprising administering the pharmaceutical composition of claim 39, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

46. A method of decreasing the expression of a protein encoded by a target RNA in a mammal comprising administering the pharmaceutical composition of claim 40, wherein the non-natural hammerhead ribozyme cleaves the target RNA.

47. A method of cleaving a target RNA sequence in a cell comprising introducing into a cell a polynucleotide comprising a nucleic acid encoding a non-natural trans-cleaving hammerhead ribozyme comprising strand A of structure I:



structure I

wherein the ribozyme cleaves a target RNA sequence comprising strand B of structure I;

wherein:

J, K, and N are each independently selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

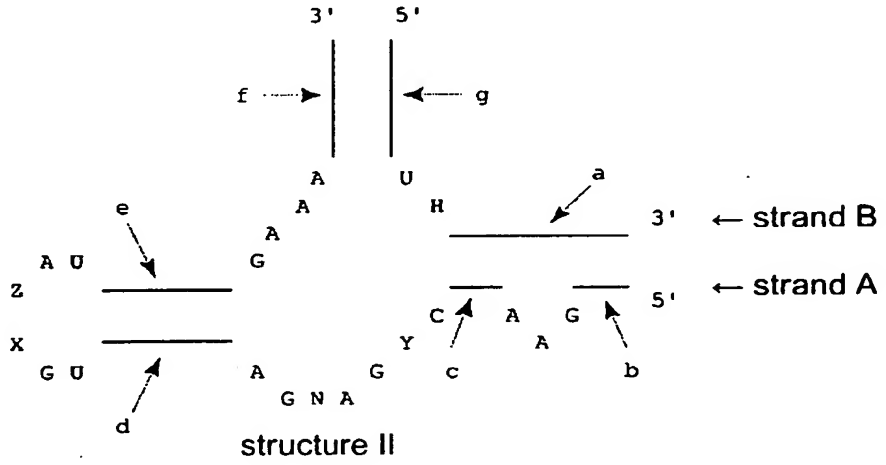
b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

a, b, JKCGUACG, and c together are stem I;

d and e together are stem II; and

f and g together are stem III.

48. A method of cleaving a target RNA sequence in a cell comprising introducing into a cell a polynucleotide comprising a nucleic acid encoding a non-natural trans-cleaving hammerhead ribozyme comprising strand A of structure II:



structure II

wherein the ribozyme cleaves a target RNA sequence comprising strand B of structure II;

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

X is selected from G and C;

Z is selected from G and A;

H is selected from A, C, and U;

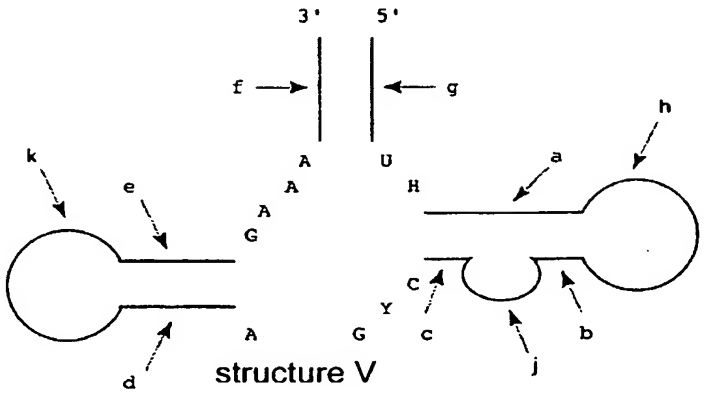
a, b, GAA, and c together are stem I;

d and e together are stem II; and

f and g together are stem III.

49. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure V:



structure V

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

h is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, and h; and

wherein each member of the library does not comprise the same j and k.

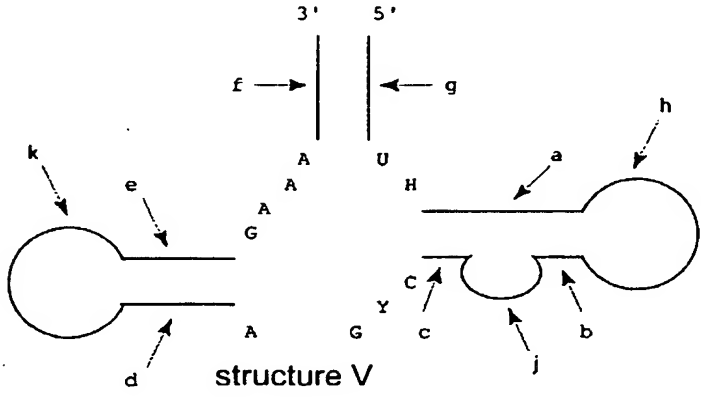
(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, f.

50. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure V:



structure V

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

h is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, h, and j; and

wherein each member of the library does not comprise the same k.

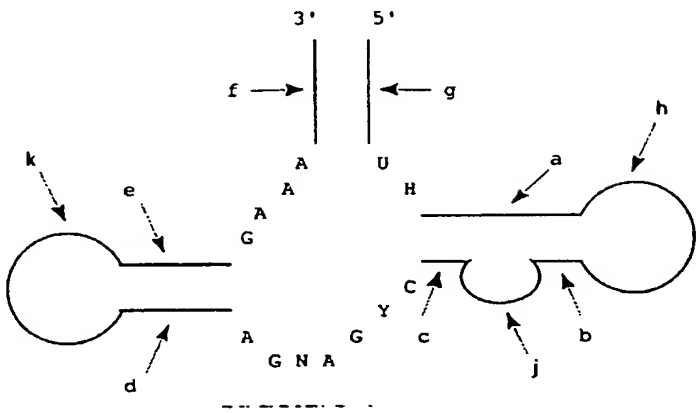
(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, and f of the selected hammerhead ribozyme.

51. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure V:



wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

h is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, h, and k; and

wherein each member of the library does not comprise the same j.

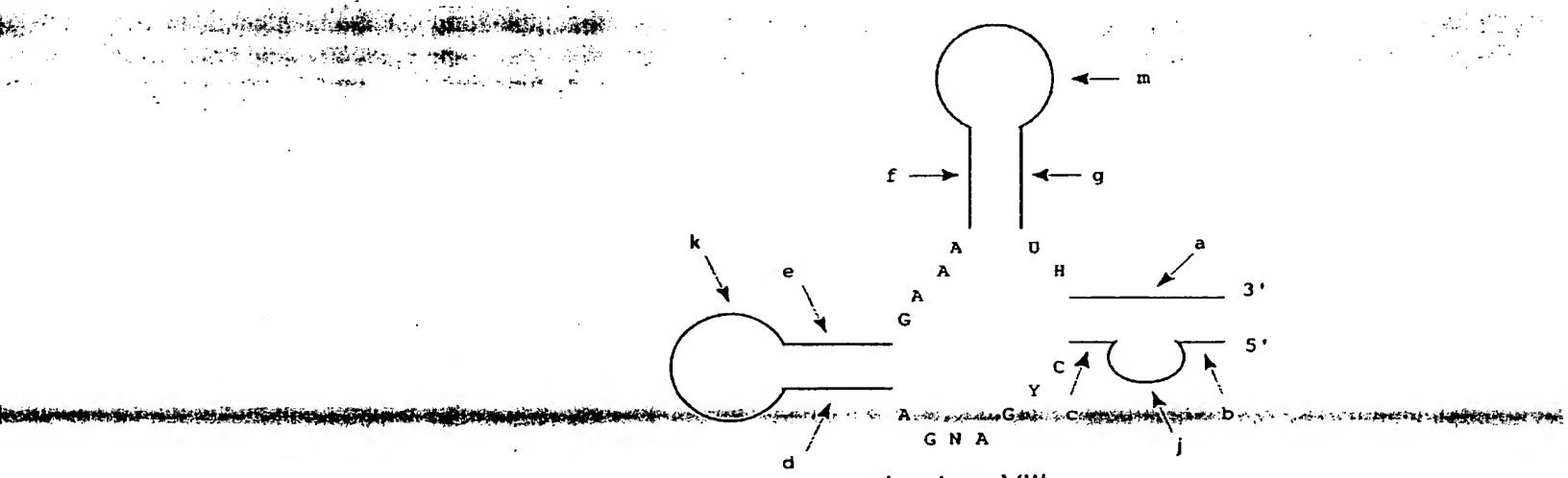
(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, and f of the selected hammerhead ribozyme.

52. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure VIII:



structure VIII

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

m is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, and h; and

wherein each member of the library does not comprise the same j and k.

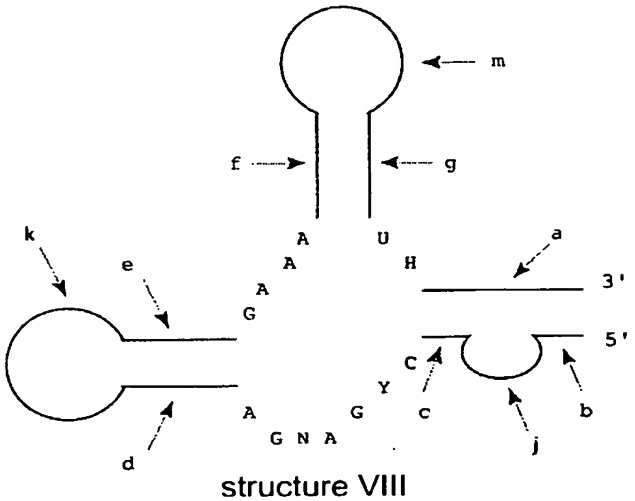
(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, f.

53. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure VIII:



structure VIII

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

m is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, h, and j; and

wherein each member of the library does not comprise the same k.

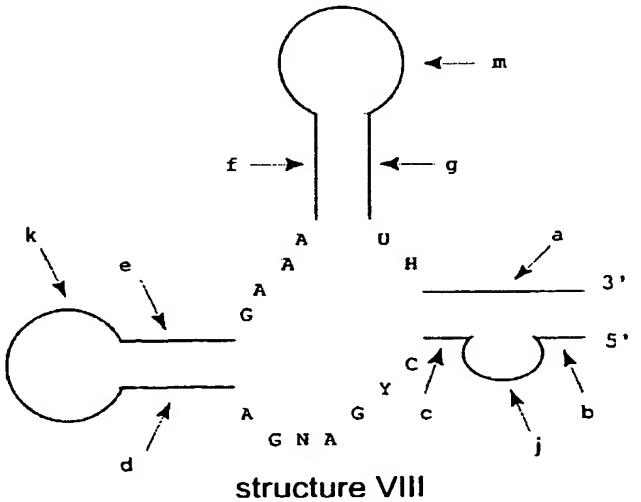
(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, and f of the selected hammerhead ribozyme.

54. A method of making a trans-cleaving hammerhead ribozyme, comprising:

(a) forming a library of hammerhead ribozyme molecules having structure VIII:



structure VIII

wherein:

N is selected from A, C, G, and U;

Y is selected from C and U;

H is selected from A, C, and U;

a is a sequence of 4-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

b, c, d, e, f, and g are each a sequence of 2-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

m is a sequence of 1-20 nucleotides, wherein each nucleotide is independently selected from A, C, G, and U;

j is a bulge of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

k is a loop of 1-20 nucleotides, wherein each nucleotide is individually selected from A, C, G, and U;

a, b, j, and c together are stem I;

d and e together are stem II;

f and g together are stem III;

wherein each member of the library comprises the same N, Y, H, a, b, c, d, e, f, g, h, and k; and

wherein each member of the library does not comprise the same j.

(b) incubating the library in the presence of less than 1 mM Mg^{2+}; and

(c) selecting a hammerhead ribozyme that cleaves at an initial rate of at least 0.5 min^{-1}.

(d) making a trans-cleaving hammerhead ribozyme comprising the nucleic acid sequence b, j, c, CYGANGA, d, k, e, GAAA, and f of the selected hammerhead ribozyme.

55. The method of any of claims 49-54, wherein the selecting comprises denaturing polyacrylamide gel electrophoresis (denaturing PAGE).

56. The method of any of claims 49-54, wherein making the trans-cleaving hammerhead ribozyme comprises transcribing a DNA that encodes the sequence b, j, c, CYGANGA, d, k, e, GAAA, f.

57. The method of any of claims 49-54, wherein making the trans-cleaving hammerhead ribozyme comprises chemically synthesizing a nucleic acid comprising the sequence b, j, c, CYGANGA, d, k, e, GAAA, f.